Irradiation of reduced-sodium uncooked lamb sausage: antimicrobial efficacy and physicochemical impact

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Abstract

Excess sodium in food is associated with an increased risk of noncommunicable diseases. Faced with this reality, consumers are becoming aware of the importance of eating foods with reduced sodium content. This work assesses the effects of partial replacement of NaCl by KCl on the microbiological and physicochemical characteristics of uncooked lamb sausage treated with different doses of gamma radiation. Analyses of thermotolerant coliforms, sulfite-reducing *Clostridium*, *Salmonella* sp., protein, moisture, ash, lipids, pH, water activity, and color were performed. The results showed that the formulations with reduced sodium content did not differ significantly concerning microbial growth and met the identity and quality standards considering its physicochemical characteristics. Also, irradiation is efficient as a microbiological control method, and the ± 3 -kGy dose was the most effective to eliminate pathogenic bacteria present in the sausage formulation prepared with reduced sodium content, inducing less intense changes in physicochemical parameters.

Keywords Ionizing radiation · Reduced salt content · Sausages

Introduction

In addition to improving flavor, sodium has other important roles in foods, especially concerning safety and characteristics like texture and consistency [1]. The World Health Organization recommends that sodium daily intake does not exceed 3 g. Despite that, sodium consumption in most

countries is above that limit [2]. Excess sodium has been linked with higher risk of noncommunicable chronic diseases (NTCD), which are responsible for 63% of deaths worldwide and 72% in Brazil [3]. In this scenario, currently, alternatives are investigated in the effort to reduce sodium levels in foods, such as the partial replacement of sodium chloride (NaCl) by potassium chloride (KCl) [4].

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Due to the increasing consumer demand for healthier, safer, and tastier food products, new, non-thermal technologies like high pressure processing, pulsed field methods, and ultraviolet and ionizing radiations have sparked the interest of researchers. However, the high doses and intensities required by these alternative treatments to inactivate bacteria may also affect the sensory and physicochemical characteristics of the end product. To face this obstacle, the combination of non-thermal and conventional methods to preserve foods may become a viable alternative to afford to use lower doses and still ensure the required levels of antimicrobial effect [5].

Food irradiation may be used to improve food safety by reducing bacterial growth, therefore improving shelf life [6]. The present study evaluated the effect of replacing NaCl by KCl combined with the use of different gamma radiation doses on microbiological and physicochemical characteristics of uncooked lamb sausage.

Materials and methods

Two uncooked lamb sausage were developed in the pilot plant of food processing of the Federal Institute of Education, Science and Technology (IFES), Campus Alegre, ES. One formulation was prepared with 2.2% NaCl, while the other was made by replacing half the NaCl content with KCl (1.1% of each salt, by weight). The formulation of the sausages can be seen in the table below (Table 1).

After preparation, sausages were packaged in polyethylene trays and surrounded by PVC film, duly labeled identifying the treatment to which they belong and frozen at -18°C . Next, sausages were transferred to a polystyrene thermal box and transported to the Laboratory of Nuclear Studies, Alberto Luiz Engineering Post-Graduation and Research Center, Federal University of Rio de Janeiro, state of Rio de Janeiro, Brazil. Subsequently, sausages were exposed to gamma radiation from a cobalt (^{60}Co) source. The doses used were ± 3 kGy and ± 5 kGy. Mean irradiation dose was 16.71 Gy/min. The dose rate in the Gammacell irradiator was

determined using Fricke dosimetry and the absolute dose by means of Silver Dichromate dosimeters screened at the National Laboratory of Ionizing Radiation Metrology (LNMRI) at the National Nuclear Energy Commission (CNEN). The samples were kept inside the irradiator by means of a cryogenic system using liquid nitrogen to maintain the temperature at around -18°C , the same temperature used to keep unirradiated samples. Sausages without NaCl replacement and not exposed to radiation were used as controls.

After irradiation, the samples were frozen at -18°C , to ensure biosafety protection and conservation, and transported to the Food Technology Laboratory, Center of Agriculture and Livestock Technologies, Universidade Estadual do Norte Fluminense Darcy Ribeiro (LTA, CCTA, UENF), where they were appropriately stored for 3 and 4 months.

Before analysis, samples were thawed at 2 to 4°C in a refrigerator for 12 h. Then, thermotolerant coliform, *Salmonella* sp., and sulfite-reducing *Clostridium* counts were carried out according to the official procedures to analyze water and meat products adopted in Brazil [7].

The physicochemical analyses were conducted in triplicate, according to official regulations and Cecchi (2003) [8, 9]. Sausage samples were analyzed for humidity, protein, ash, lipid levels, pH, water activity (a_w), and color.

The data obtained were submitted to statistical analysis based on completely randomized design and analysis of variance (ANOVA) and the Tukey and chi-square tests in the SAS software version 9.3 [10].

Results

There was no coliform growth or gas production at 45°C in uncooked lamb sausages formulated with 100% NaCl or 50% NaCl and 50% KCl and submitted to radiation doses of ± 3 kGy and ± 5 kGy for 3 and 4 months of storage under freezing. Under these same conditions, there was also no growth of sulfite-reducing *Clostridium*.

Similarly, no *Salmonella* sp. growth was detected in uncooked lamb sausage prepared with 50% NaCl and 50% KCl and in those made with 100% NaCl treated with ± 3 kGy and ± 5 kGy 3 and 4 months into storage. However, non-irradiated samples were contaminated with the bacterium irrespective of the formulation (50% NaCl and 50% KCl, and 100% NaCl).

The presence of enterobacteria in non-irradiated samples was confirmed by serum culture. Statistically significant differences were observed in *Salmonella* sp. levels between non-irradiated lamb sausages and the samples irradiated with both ± 3 kGy and ± 5 kGy (chi-square test, $p > 0.001$). These results afford to conclude that the ± 3 -kGy gamma radiation dose is high enough to eliminate enterobacteria in uncooked lamb sausage.

Table 1 Formulation of the sausages given in percentage of ingredients

	1 formulation (100% NaCl)	2 formulation (50% NaCl e 50% KCl)
Meat	75.66%	75.66%
Fat	20.00%	20.00%
NaCl	2.20%	1.10%
KCl	0.00%	1.10%
Sugar	0.095%	0.095%
H ₂ O	2.00%	2.00%
Nitrite	0.015%	0.015%
Erythorbate	0.025%	0.025%

The partial replacement of NaCl levels by KCl did not affect pH, humidity, and a_w or protein, lipid, and ash levels significantly ($p > 0.05$). Yet, the lamb sausage formulation using KCl to replace 50% of the NaCl content met the identity and quality standards (IQS), which are part of official Brazilian regulations applicable (Table 2) [12].

The gamma radiation doses used in the present study (± 3 kGy and ± 5 kGy) influenced pH of uncooked lamb sausage. Significant differences were observed ($p > 0.05$) between sausages treated with the two doses. In addition, the values varied significantly ($p > 0.05$) between the formulations with two salt levels treated with both radiation doses, indicating the interaction between the variables. The pH values of irradiated sausages were lower compared with those of non-irradiated sausages.

The results of the present study indicate how a_w and lipid content are influenced by the interaction ($p > 0.05$) between the partial replacement of NaCl and the ± 5 -kGy dose. Importantly, a_w was significantly higher in the low-sodium formulation (50% NaCl, 50% KCl) treated with ± 5 kGy, with significant difference ($p > 0.05$) when compared with the non-irradiated control and the sausages treated with ± 3 kGy and prepared with the same salt composition.

Lipid levels were also higher in the low-sodium formulation (50% NaCl and 50% KCl) treated with ± 5 kGy, with significant difference ($p > 0.05$) when compared with the control and the ± 3 kGy formulations.

Tables 2 and 3 show the means, standard deviations, and results of analysis of variance obtained using the mathematical models coded for luminosity (L^*) and chroma values (C^*) for one non-irradiated control formulation and two irradiated uncooked lamb sausage formulations prepared with two salt concentrations. No variable significantly affected L^* ($p < 0.05$) (Table 4).

Table 3 Mean and standard deviation (SD) of color of uncooked lamb sausage for the two salt concentrations used

Color	L^*		C^*	
	Mean	SD	Mean	SD
Salt levels				
100% NaCl	46.20a	± 1.42	14.58a	± 0.74
50% NaCl 50% KCl	46.36a	± 1.87	15.71b	± 1.22

Means followed by different lowercase letters in columns differ statistically concerning color for the two salt concentrations at 5% probability in the Tukey test

L^* of sausages prepared with KCl replacing 50% of the NaCl content did not vary significantly, compared with the formulation prepared with NaCl alone ($p < 0.05$). However, C^* values were significantly higher ($p > 0.05$) in the 50% NaCl + 50% KCl formulation, indicating color saturation. No statistically significant difference was observed for the interaction between treatments ($p > 0.05$).

Discussion

The results of the present study confirm the findings published by Oliveira et al. [13], the authors found that doses of 1.5 kGy and 3 kGy were adequate to eliminate *Salmonella* sp. from chicken breast. Similar results were also observed by Henriques et al. [14], who treated lamb meat with radiation doses of ± 3 kGy and ± 5 kGy and reported that the low dose used is sufficient to eliminate *Salmonella* sp.

Our results also show the importance of using radiation as a food safety measure, since the presence of salmonella in foods is a serious public health problem. It has been shown that signs

Table 2 Mean and standard deviation (SD) of physicochemical parameters of treatments and identity and quality standards (IQS) of uncooked sausage

Treatments	Humidity %	Protein %	Lipids %	Ash %	Water activity	pH
Non-irradiated, 100% NaCl	62.77a ± 1.99	13.42a ± 4.30	20.04a ± 1.13	2.79b ± 0.08	0.95a ± 0.006	6.33a ± 0.10
Non-irradiated, 50% de NaCl	63.36a ± 0.37	13.38a ± 0.33	18.89b ± 0.70	2.76c ± 0.10	0.94a ± 0.006	6.61b ± 0.15
Irradiated with 3 kGy, 100% NaCl	61.60a ± 1.00	13.87a ± 1.16	18.61a ± 0.45	3.11a ± 0.07	0.94b ± 0.009	5.99a ± 0.03
Irradiated with 3 kGy, 50% NaCl	63.25a ± 0.61	15.88a ± 3.65	19.47b ± 0.75	2.97b ± 0.12	0.95b ± 0.002	6.07b ± 0.01
Irradiated with 5 kGy, 100% NaCl	63.34a ± 1.36	12.88a ± 1.77	19.80a ± 1.11	2.80c ± 0.06	0.95b ± 0.005	5.97a ± 0.05
Irradiated with 5 kGy, 50% NaCl	59.58a ± 2.69	17.41a ± 2.61	22.14b ± 1.64	3.28a ± 0.06	0.97b ± 0.001	5.96a ± 0.02
IQS [§]	≤ 70	≥ 12	≤ 30			

Means followed by different lowercase letters in columns differ statistically concerning irradiation at 5% probability in the Tukey test

[§] Brasil [11]

Table 4 Mean and standard deviation (SD) of color of uncooked lamb sausage for the two radiation doses used

Color	L*		C*	
	Mean	SD	Mean	SD
Radiation dose				
Control	46.38a	± 2.27	15.52a	± 1.58
3 kGy	46.05a	± 1.47	15.08a	± 0.91
5 kGy	46.39a	± 1.09	14.84a	± 0.79

Means followed by different lowercase letters in columns differ statistically concerning color for the two radiation doses at 5% probability in the Tukey test

and symptoms of salmonellosis may be misdiagnosed, ultimately overloading health systems [11].

Concerning the effect of radiation on ash content, other authors did not observe any effect of gamma radiation on ash contents of fish and mollusk [15, 16]. This contrasts with the results of the present study, when significant differences were observed in ash content after use of the two radiation doses. For pH, the results obtained were similar to the findings published by Ham et al. [17], who also observed a significant interaction between irradiation and pH values in skin encased meat products. The pH values of cooked pork sausage decreased with irradiation, though the difference in pH across sausages irradiated with the doses used was under 0.1 unit, leading the authors to conclude that it may be too small to affect the quality of processed meat products. For Baptista et al. [18], pH may be influenced by the growth of enterobacteria in non-irradiated samples, when alkaline compounds are produced, increasing pH values in comparison with irradiated samples.

Dutra et al. [19] obtained similar results in a study that assessed the effect of different nitrite and radiation doses on mortadella formulations. The authors observed that pH, humidity, total protein, lipid, and ash levels of mortadella formulations were not affected ($p > 0.05$) by nitrite or radiation. However, they reported the interaction between radiation and nitrite levels.

Lipids are rather sensitive to radiation, whose direct result is the production of excited cations and anions. Subsequent reactions generate several other compounds, including diglycerides and triglycerides (esters, ketones, and hydrocarbons) [20]. These reactions may induce the rise in lipid levels in samples of low-sodium uncooked lamb sausage irradiated with ± 5 -kGy doses.

For L*, the results obtained in the present study were similar to the findings published by Kwon et al. [21], in a study about the effect of a ± 5 kGy radiation dose on frankfurters. The authors observed no significant effect of the dose used on L* values.

The objective evaluation of color is an important source of information about the quality of meat products in general.

Nascimento et al. [22], evaluated the color of low-sodium sausages with the replacement of up to 50% of the salt by KCl in formulations. The authors observed that low-sodium formulations had significantly higher L* values and significantly lower red color coordinates (a*), compared with control. In turn, coordinate b*, which measures yellow intensity, was not affected by low-sodium contents in formulations.

Conclusions

1. Gamma radiation is an efficient measure in the microbiological control of low-sodium uncooked lamb sausage.
2. A ± 3 -kGy dose is more efficient to eliminate pathogenic bacteria in low-sodium uncooked lamb sausage at the same time that it induces smaller changes in physico-chemical parameters, compared with a ± 5 -kGy dose.

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